

## CLAIMS

*sub A2*  
We Claim:

1. A reversible physiological process for the temporal separation of oxygen evolution and hydrogen production in a microorganism, comprising:

- 5 (a) growing a culture of the microorganism in medium under illuminated conditions to accumulate an endogenous substrate;
- (b) depleting from the medium a nutrient selected from the group consisting of sulfur, iron, and/or manganese;
- 10 (c) sealing the culture from atmospheric oxygen;
- (d) incubating the culture in light whereby a rate of light-induced oxygen production is equal to or less than a rate of respiration; and
- (e) collecting an evolved gas.

2. The process of claim 1 further comprising generating hydrogen from water and the accumulated substrate using light and a hydrogenase.

15 3. The process of claim 1 wherein depleting is to a concentration of 0.5 millimolar or less.

4. The process of ~~claim~~ 1 further comprising replacing a head gas with an inert gas.

5. The process of claim 1 further comprising, after incubating and collecting, *L A B* repeating the steps of growing to accumulate additional substrate, *2 A B* depleting, sealing and incubating for a plurality of cycles.

*sub A3*  
6. The process of claim 2 wherein the microorganism is selected from the group consisting of a green, red, brown, and blue-green algae.

25 7. The process of claim 2 further comprising providing the medium with the depleted nutrient after generating, and repeating the steps of growing, depleting, incubating and generating. *L A B*

8. The process of claim 2 wherein the <sup>*ind. source ?*</sup>substrate is selected from the group consisting of acetate, carbohydrate, lipid and protein.

9. The process of ~~claim~~ 4 wherein the inert gas is nitrogen.

30 10. The process of claim 6 wherein the algae is *Chlamydomonas reinhardtii*.  
*sub A4*